Please amend claims 4-5, 8, 11, 12 and 28 by changing their dependency to depend from claim 20 instead of claim 1.

Sub B1

2. (Amended) The method of claim [1] 20, further comprising [isolating an adenoviral particle] purifying adenovirus from said cell lysate by a process that includes one or more [using] chromatography steps.

A1

~~3.~~ 9 (Amended) The method of claim [1] ~~20~~ 1, wherein [the glucose concentration in said media is maintained between about 0.7 and about 1.7 g/L] the cells are perfused with a glucose containing media at a rate to provide a glucose concentration of between about 0.7 and 1.7 g/L.

Sub B2

20. (Amended) A method for producing a pharmaceutically acceptable adenovirus composition comprising:

A2

a) growing host cells in a media;

b) perfusing said host cells;

c) infecting said host cells with an adenovirus;

d) lysing said host cells to provide a cell lysate comprising adenovirus [The method of claim 1], wherein said lysis is achieved through autolysis of infected cells; and

e) purifying adenovirus from said lysate to provide a pharmaceutically acceptable adenovirus composition.

22. (Amended) The method of claim 2, wherein [said isolating consists essentially of a single chromatography step] the chromatography step comprises essentially a single chromatography step.

~~53.~~ 26 (Amended) The method of claim ~~20~~ 1 [52], wherein said media is a serum-free media and said host cells are [adapted for growth] capable of growing in serum-free media.

Please amend the dependency of claims 54 and 55 to depend from claim 20 instead of from claim 52.

Please add the following new claims, claims 70 - 117:

-- 70. A method for producing an adenovirus composition comprising:

a) growing host cells in a media comprising glucose;

b) perfusing said cells at a rate to provide a glucose concentration of less than 2.0 g/L;

c) infecting said host cells with an adenovirus; and

d) harvesting and lysing said host cells to produce a lysate comprising said adenovirus composition.

71. The method of claim 70, wherein the cells are perfused at a rate to provide a glucose concentration of less between about 0.7 and 1.7 g/L.

~~72.~~ 32 The method of claim ~~70~~, 30 wherein said adenovirus comprises an adenoviral vector comprising an exogenous gene construct.

~~73.~~ 33 The method of claim ~~72~~, 32 wherein said gene construct is operatively linked to a promoter.

~~74.~~ 34 The method of claim ~~73~~, 33 wherein said promoter is SV40 IE, RSV LTR, β-actin, CMV IE, adenovirus major late, polyoma F9-1, or tyrosinase.

~~75.~~ 38 The method of claim ~~70~~, 30 wherein said adenovirus is a replication-incompetent adenovirus.

~~76.~~ 39 The method of claim ~~75~~, 38 wherein the adenovirus is lacking at least a portion of the E1-region.

~~77.~~ 40 The method of claim ~~76~~, 39 wherein the adenovirus is lacking at least a portion of the E1A and/or E1B region.

~~78.~~ 41 The method of claim ~~70~~, 30 wherein said host cells are capable of complementing replication.

~~79.~~ 42 The method of claim ~~70~~, 30 wherein said host cells are 293 cells.

~~80.~~ 35 The method of claim ~~72~~ 32, wherein said exogenous gene construct encodes a therapeutic gene.

~~81.~~ 36 The method of claim ~~80~~ 35, wherein said therapeutic gene encodes antisense *ras*, antisense *myc*, antisense *raf*, antisense *erb*, antisense *src*, antisense *fms*, antisense *jun*, antisense *trk*, antisense *ret*, antisense *gsp*, antisense *hst*, antisense *bcl* antisense *abl*, Rb, CFTR, p16, p21, p27, p57, p73, C-CAM, APC, CTS-1, zac1, scFV *ras*, DCC, NF-1, NF-2, WT-1, MEN-I, MEN-II, BRCA1, VHL, MMAC1, FCC, MCC, BRCA2, IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11 IL-12, GM-CSF G-CSF, thymidine kinase or p53.

~~82.~~ 37 The method of claim ~~81~~ 36, wherein said therapeutic gene encodes p53.

A5 Cont

~~83.~~ 43 The method of claim ~~70~~ 30, wherein said cells are harvested and lysed *ex situ* using a hypotonic solution, hypertonic solution, freeze-thaw, sonication, impinging jet, microfluidization or a detergent.

~~84.~~ 44 The method of claim ~~70~~ 30, wherein said cells are harvested and lysed *in situ* using a hypotonic solution, hypertonic solution, or a detergent.

~~85.~~ 45 The method of claim ~~84~~ 44, wherein said cells are lysed and harvested using detergent.

~~86.~~ 46 The method of claim ~~85~~ 45, wherein said detergent is Thesit®, NP-40®, Tween-20®, Brij-58®, Triton X®-100 or octyl glucoside.

~~87.~~ 47 The method of claim ~~70~~ 30, further comprising purifying adenovirus from said cell lysate.

~~88.~~ 57 The method of claim ~~70~~ 30, wherein said media is subjected to diafiltration.

~~89.~~ 48 The method of claim ~~87~~ 47, wherein said purifying involves treatment of the cell lysate with a nuclease.

~~90.~~ 49 The method of claim ~~89~~ 48, wherein said nuclease is Benzonase® or Pulmozyme®.

AS Unt

~~91.~~ 50 The method of claim ~~87~~ 47, wherein said purifying includes chromatography using one or more chromatography steps.

SUB B6

92. The method of claim 91, wherein said chromatography comprises essentially a single chromatography step.

~~93.~~ 52 The method of claim ~~92~~ 51, wherein said chromatography step involves ion exchange chromatography.

~~94.~~ 53 The method of claim ~~93~~, 52 wherein said ion exchange chromatography is anion exchange chromatography.

~~95.~~ 54 The method of claim ~~94~~, 53 wherein said anion exchange chromatography utilizes DEAE, TMAE, QAE, or PEI.

~~96.~~ 55 The method of claim ~~95~~, 54 wherein said anion exchange chromatography utilizes Toyopearl Super Q 650M, MonoQ, Source Q or Fractogel TMAE.

~~97.~~ 56 The method of claim ~~94~~, 55 wherein said ion exchange chromatography is carried out at a pH range of between about 7.0 and about 10.0.

~~98.~~ 58 The method of claim ~~70~~, 30 further comprising a concentration step employing membrane filtration.

~~99.~~ 59 The method of claim ~~98~~, 58 wherein said filtration is tangential flow filtration.

~~100.~~ 60 The method of claim ~~98~~, 58 wherein said filtration utilizes a 100 to 300K NMWC, regenerated cellulose, or polyether sulfone membrane.

Sub B7 101. A method for preparing a pharmaceutically acceptable adenovirus composition comprising:

a) growing host cells;

b) infecting said host cells with an adenovirus;

c) lysing said host cells using a lysing technique other than freeze-thaw to produce a crude lysate composition comprising adenovirus; and

d) purifying adenovirus from said lysate by a process that includes one or more chromatography steps, to provide a pharmaceutically acceptable adenovirus composition.

~~102.~~ 62 The method of claim ~~101~~, 61 wherein the cells are lysed by solid shear, detergent lysis, liquid shear or sonication, to produce a cell lysate.

~~103.~~ 63 The method of claim ~~102~~, 62 wherein the cells are lysed by detergent lysis.

~~104.~~ 64 The method of claim ~~103~~, 63 wherein the cells are lysed by detergent Thesit®, NP-40®, Tween-20®, Brij-58®, Triton X-100® or octyl glucoside.

~~105.~~ 65 The method of claim ~~104~~, 64 wherein said detergent is present in the lysis solution at a concentration of about 1% (w/v).

~~106.~~ 66 The method of claim ~~101~~, 61 wherein the cells are perfused in a glucose containing media at a rate to provide a glucose concentration of between about 0.7 and 1.7 g/L.

~~107.~~ 67 The method of claim ~~101~~ 61, wherein said lysate is subjected to a diafiltration step.

108. The method of claim 101, wherein the chromatography comprises essentially a single chromatography step.

~~109.~~ 69 The method of claim ~~108~~ 68, wherein said chromatography step involves ion exchange chromatography.

110. A method for preparing a pharmaceutically acceptable adenovirus composition comprising:

a) growing host cells in a media;

b) infecting said host cells with an adenovirus; and

c) harvesting and lysing said host cells to provide a lysate comprising adenovirus; and

d) purifying adenovirus from said lysate by a process that includes a chromatography step, wherein said chromatography step involves essentially a single chromatography step, to provide a pharmaceutically acceptable adenovirus composition.

111. The method of claim 110, wherein said chromatography step involves ion exchange chromatography.

112. The method of claim 111, wherein said ion exchange chromatography is anion exchange chromatography.

113. The method of claim 112, wherein said anion exchange chromatography utilizes DEAE, TMAE, QAE, or PEI.

114. The method of claim 112, wherein said anion exchange chromatography utilizes Toyopearl Super Q 650M, MonoQ, Source Q or Fractogel TMAE.

115. The method of claim 110, wherein the adenovirus is subjected to purification steps that include reducing the concentration of contaminating nucleic acid.

116. The method of claim 110, further defined as comprising the steps of concentrating said crude cell lysate, exchanging buffer of said crude cell lysate, and reducing the concentration of contaminating nucleic acids in said crude cell lysate.

117. The method of claim 110, wherein said media is serum-free media and the cells are capable of growing in serum-free media.

118. A method for preparing a pharmaceutically acceptable adenovirus composition comprising:

a) growing host cells;

b) perfusing said host cells;

c) infecting said host cells with an adenovirus;

d) lysing said host cells using a lysing technique other than freeze-thaw to produce a crude lysate composition comprising adenovirus; and

e) purifying adenovirus from said lysate by a process that includes a chromatography step, wherein said chromatography step involves essentially a single chromatography step, to provide a pharmaceutically acceptable adenovirus composition.

119. The method of claim 20, 70 or 118, wherein the perfusion is achieved by a fed-batch process.

120. The method of claim 20, 70 or 118, wherein the perfusion is achieved by continuous perfusion--

REMARKS

I. Claims in the Case, and Amendments to the Claims

Claims 1, 16-19, 31-52, 57-69 have been canceled without prejudice to represent the subject matter of these claims in later continuing cases. Claims 2-5, 8, 11, 12, 20, 22, 28, 53-55 have been amended, and claims 70-120 have been added. Claims 2-15, 20-30, 53-56 and 70-120 are currently pending.